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**Figure 1A: CTLA-4 VLD-SPECIFIC OLIGONUCLEOTIDES**

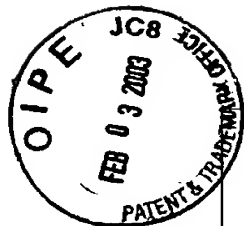
OLIGONUCLEOTIDE	NUMBER	SIZE	SEQUENCE <sup>1</sup>	FEATURES
For 5' CTLA-4 amplification	3553 5023 5671	54	TTATTACTGCGGCCAGCCGGCCATggCCgCAATgCACgTggCC CAGCCTgCT (SEQ ID NO:2)	forward primer + SfiI site
For 5' CTLA-4 amplification	5445	60	TTATTACTGCGGCCAGCCGGCCATggCCgCAATgCACgTggCC CAGCCTgCTgTggTA (SEQ ID NO:3)	forward primer + SfiI site
For 5' CTLA-4 amplification	5443	45	TCTCACAgTgCACAggCAATgCACgTggCCCCAgCCTgCTgTggTA (SEQ ID NO:4)	forward primer + ApaLI site
For 5' CTLA-4 amplification	4851	39	TCTCACAgTgCACAggCAATgCACgTggCCCCAgCCTgCT (SEQ ID NO:5)	forward primer + ApaLI site
For 5' CTLA-4 amplification	5467	43	gCCCAGCCggCCgAAATTCgCAATgCACgTggCCCCAgCCTgCTg (SEQ ID NO:6)	forward primer + EcoRI site
For 5' CTLA-4 amplification		60	GCAGCTAATACGACTCACTATAGGAACAGACCACCATGGA CGTGGCCCAGCCTGCTGTGG (SEQ ID NO:7)	ribosomal display
For 3' CTLA-4 amplification	4316 5022 5670	42	ATCTgCggCCgCTACATAAAATCTgggTACCgTTgCCgATgCC (SEQ ID NO:8)	reverse primer + NotI site
For 3' CTLA-4 amplification	4486	66	gCTgAAATTCtATCAGTgATggTgATggTgATgTgCggCCgCgTCag AATCTgggCACggTTCtgg (SEQ ID NO:9)	
For 3' CTLA-4 amplification		51	GCCCTTGGGCGGGAGATGGTCTGTCTCAGTGGCGAGGGC AGGTCTGTGTG (SEQ ID NO:10)	ribosomal display 1
For 3' CTLA-4 amplification		49	CGAGGGCAGGTCTGTGTGGGTCACGGTGCACGTGAACCTCT CCCCGGAG (SEQ ID NO:11)	ribosomal display 2

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**Figure 1B: CTLA-4 VLD-SPECIFIC OLIGONUCLEOTIDES**

OLIGONUCLEOTIDE	NUMBER	SIZE	SEQUENCE	FEATURES
For 3' CTLA-4 amplification		51	CGTGAACCTCTCCCGGAGTTCCAGTCATCCTCGCAGATGC TGGCCTCACC (SEQ ID NO:12)	ribosomal display 3
For CDR1-somatostatin	4585	84	AgCTTTgTgTgAgTATgCagCTggCTgCAAgAATTCTTCTTgA AgACTTTTCACATCCTTgTgCCACTgAggTCCgggTgACA (SEQ ID NO:13)	forward primer + Som
For CDR3-somatostatin	4586	84	CTgggTACCgTTCCgATgCCACAggATgTgAAAgTCTTCCAgAA gAAATTCTTgCagCCAgCCTCCACCTTgCAGATgTAgAg (SEQ ID NO:14)	reverse primer + Som
For CDR1-som-randomisation	4835	75	AgCTTTgTgTgAgTATgCagCTggCTgCAAgAATNNg/TNNg/T NNg/TNNg/TNNg/TNNg/TACATCCTTgTgCCACTgAggTC (SEQ ID NO:15)	
For CDR3-som-randomisation	4836	75	CTgggTACCgTTCCgATgCCACAggATgTA/CNNA/CNNA/CNN A/CNNA/CNNA/CNNAATTCTTgCagCCAgCCTCCACCTTgCA (SEQ ID NO:16)	
For CDR2 haemagglutinin tag	4766	21	gTAggTTgCCgCACAgACTTC (SEQ ID NO:17)	back primer for splice overlap
For CDR2 haemagglutinin tag	4775	66	gAAGTCTgTgCggCAACCTACCCgTATgACgTTCCggACTACgCC CTAgATgATTCCATCTgCACg (SEQ ID NO:18)	forward primer tag overlap
For CDR-1 anti-lysozyme	5232	78	gCCAgCTTTgTgTgTgAgTATgCCAgTggCTACACCATCgggCCgT ACTgCATgggCgTCCgggTgACAgTgCTTCgg (SEQ ID NO:19)	
For CDR-2 anti-lysozyme	5228	60	TgTgCggCagCCATCAACATgggCggTggCATCACCTTCCTAgAT gATTCCATCTgCACg (SEQ ID NO:20)	Forward



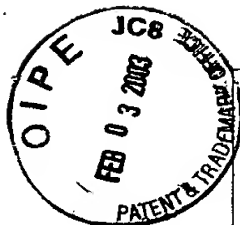
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**Figure 1D: CTLA-4 VLD-SPECIFIC OLIGONUCLEOTIDES**

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**Figure 1E: CTLA-4 VLD-SPECIFIC OLIGONUCLEOTIDES**

OLIGONUCLEOTIDE	NUMBER	SIZE	SEQUENCE	FEATURES
CDR3 Randomisation	5470	78	AATCTgggTACCCgTTgCCgATgCC( <sup>Δ</sup> /cNN) <sub>10</sub> CACCTTgCAGATgTAgATCCCgT (SEQ ID NO:43)	
CDR3 Randomisation	5474	93	AATCTgggTACCCgTTgCCgATgCCCAg( <sup>Δ</sup> /cNN) <sub>13</sub> CTCCACCTTgCAGATgTAgATCCCgT (SEQ ID NO:44)	
CDR3 Randomisation	5471	81	AATCTgggTACCCgTTgCCgATgCC( <sup>Δ</sup> /cNN) <sub>4</sub> gCA( <sup>Δ</sup> /cNN) <sub>6</sub> CACCTTgCAGATgTAgATCCCgT (SEQ ID NO:45)	
CDR3 Randomisation	5472	87	AATCTgggTACCCgTTgCCgATgCC( <sup>Δ</sup> /cNN) <sub>3</sub> gCA( <sup>Δ</sup> /cNN) <sub>7</sub> CACCTTgCAGATgTAgATCCCgT (SEQ ID NO:46)	
CDR3 Randomisation	5475	99	AATCTgggTACCCgTTgCCgATgCC( <sup>Δ</sup> /cNN) <sub>6</sub> gCA( <sup>Δ</sup> /cNN) <sub>10</sub> CACCTTgCAGATgTAgATCCCgT (SEQ ID NO:47)	
CDR3 Randomisation	5473	87	AATCTgggTACCCgTTgCCgATgCC( <sup>Δ</sup> /cNN) <sub>4</sub> gCA( <sup>Δ</sup> /cNN) <sub>7</sub> CACACCTTgCAGATgTAgATCCCgT (SEQ ID NO:48)	
CTLA-4 codon-change	5591	70	ATgCACgTggCCCCAgCCTgTgTggTgCTggCCA gCgCCgTggCATgCgCgCTTgTgTgAAATATg (SEQ ID NO:49)	
CTLA-4 codon-change	5592	77	gCCAgCTTTgTgTgTgAAATATgCgTCTggCTATACCATCgCCCgTACTgCATgggTgCgTgTgAACgTgCTgCg (SEQ ID NO:50)	
CTLA-4 codon-change	5598	54	gTgCgTgTgACCgTgCTgCgTCAggCggATAgCCAggTgACCgAAgTTTgCgCg (SEQ ID NO:51)	
CTLA-4 codon-change	5600	75	CAGgTgACCgAAgTTTgCgCgCgCgCgATCAACATgggCggTggCATCACCTTCTgTgTgATTCATCTgCACC (SEQ ID NO:52)	
CTLA-4 codon-change	5599	66	CAGACCCCTggATggTCAggTTCACCTggTTACCCgCTggAggTgCCgTgCAGATggAAATCATCCAg (SEQ ID NO:53)	
CTLA-4 codon-change	5597	57	CACTTTgCAGATgTACAgACCggTATCCATggCACgCgAACCTTggATggTCAggTT (SEQ ID NO:54)	

ALL INFORMATION CONTAINED HEREIN IS UNCLASSIFIED

<sup>1</sup> All oligonucleotides are described 5' to 3'.  
N represents combination of the four nucleotides.

<sup>1</sup> All oligonucleotides are described 5' to 3'.

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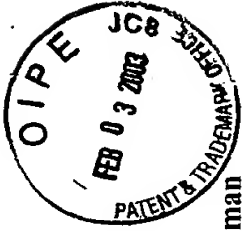


Figure 2: Polynucleotide sequence of complete cDNA encoding human CTLA-4 and polypeptide sequence of the VLD of human CTLA-4.

A. Polynucleotide Encoding Human CTLA-4 cDNA (SEQ ID NO:57)

```
ATGGCTTGCC TTGGATTTC ACGGCACAAG GCTCAGCTGA ACCTGGCTGC CAGGACCTGG CCCTGCACTC TCCTGTTTTT... 80
TCTTCTCTTC ATCCCTGTCT TCTGCAAAAGC AATGCACGTG GCCCAGCCTG CTGTGGTACT GGCCAGCAGC CGAGGCATCG... 160
CCAGCTTTGT GTGTGAGTAT GCATCTCCAG GCAAAGCCAC TGAGGTCCGG GTGACACATGC TTCCGGCAGGC TGACAGCCAG... 240
GTGACTGAAG TCTGTGCGGC AACCTACATG ACGGGGAATG AGTTGACCTT CCTAGATGAT TCCATCTGCA CGGGCACCTC... 320
CAGTGGAAAT CAAGTGAACC TCACTATCCA AGGACTGAGG GCCATGGACA CGGGACTCTA CATCTGCAAG GTGGAGCTCA... 400
TGTACCCACC GCCATACTAC CTGGGCATAG GCAACGGAAC CCAGATTAT GTAATTGATC CAGAACCCTG CCCAGATTCT... 480
GACTTCCTCC TCTGGATCCT TGCAGCAGTT AGTTCGGGGT TGTTTTTTA TAGCTTTCTC CTCACAGCTG TTCTCTTGAG... 560
CAAAATGCTA AAGAAAAGAA GCCCTCTTAC AACAGGGGTC TATGTGAAAA TGCCCCCAAC AGAGCCAGAA TGTGAAAAGC... 640
AATTCAGCC TTATTTTATT CCCATCAATT GA ... 672
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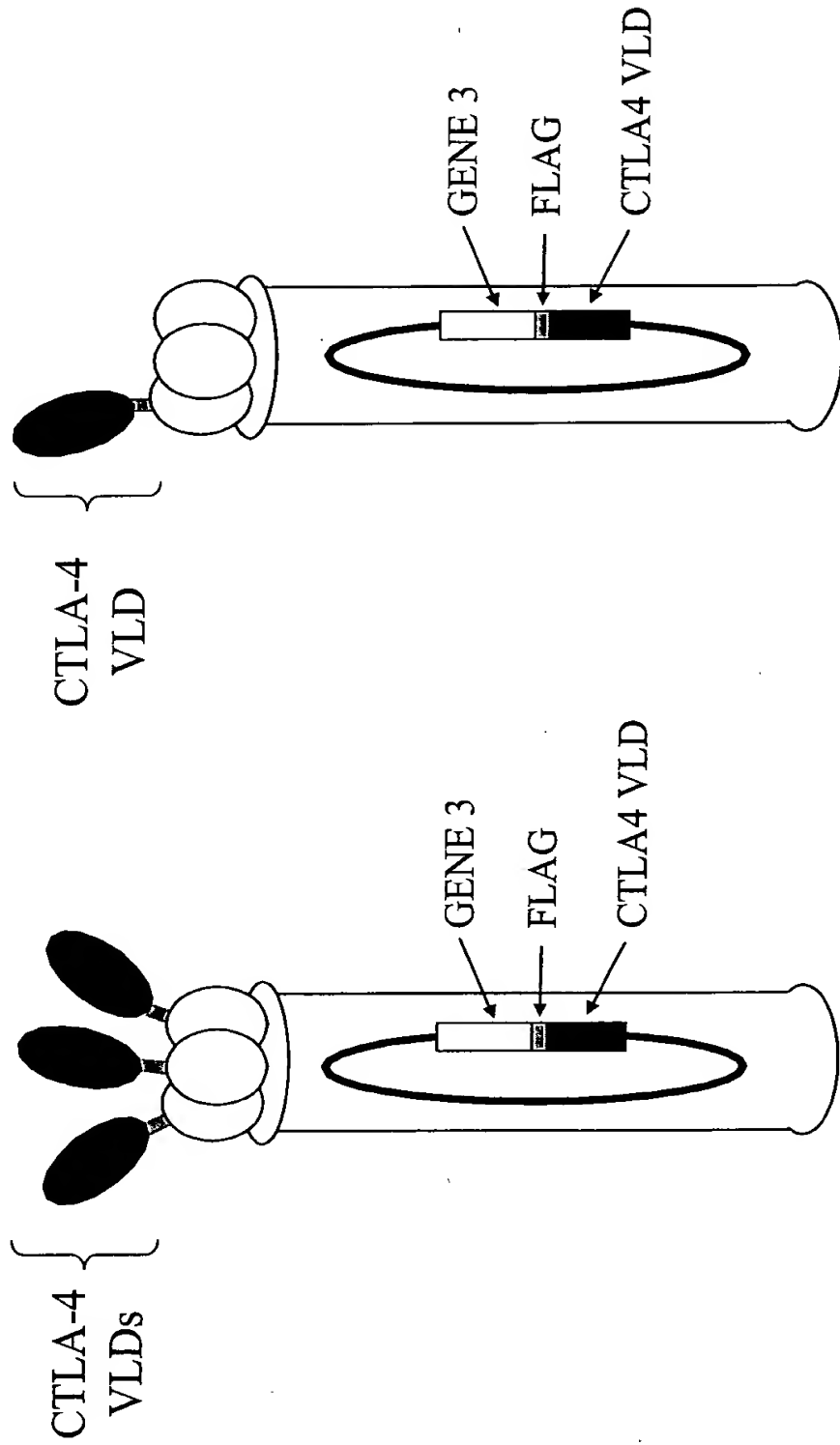
B. Polypeptide Sequence of Human CTLA-4 VLD (SEQ ID NO:58)

```
1 10 20 30 40 50 60 70 80
AMHVAQPAV LASSRGIAF VCEYASPGK TEVRVTVLRQ ADSQVTEVCA ATYMMGNELT FLDDSICTGT SSGNQVNLT
QGLRAMDTGL YICKVELMYP PPYILGIGNG AQIYV
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Figure 3: Phage display of CTLA-4 VLD STMs

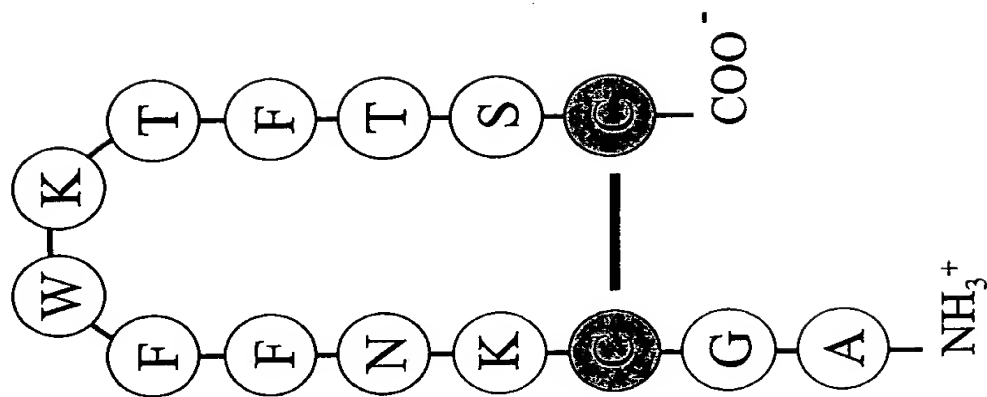




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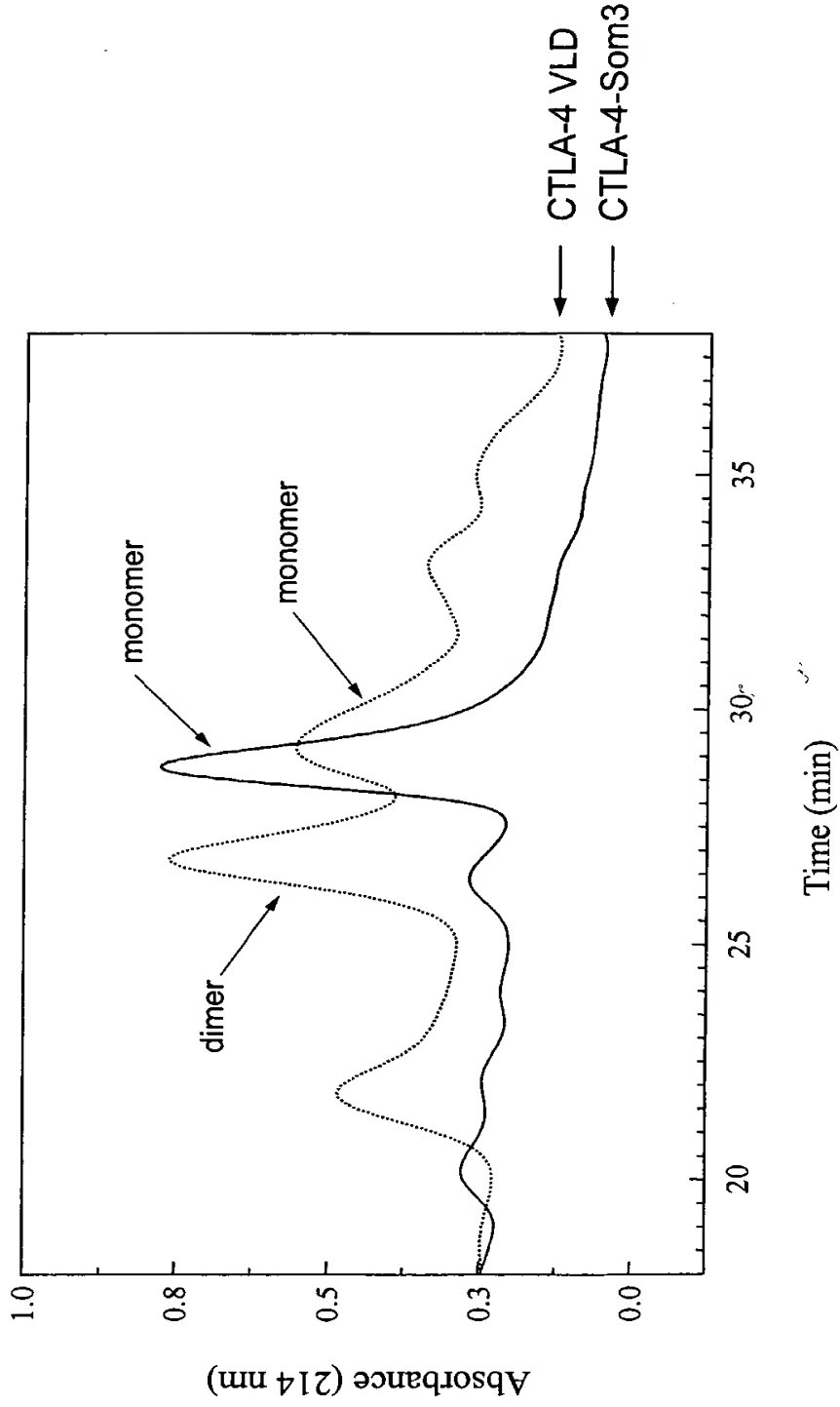
Figure 4: Schematic representation of the somatostatin peptide. (SEQ ID NO:139)



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Figure 5: Comparison of the HPLC profiles of affinity purified CTLA-4 VLD and CTLA-4-Som3 STM





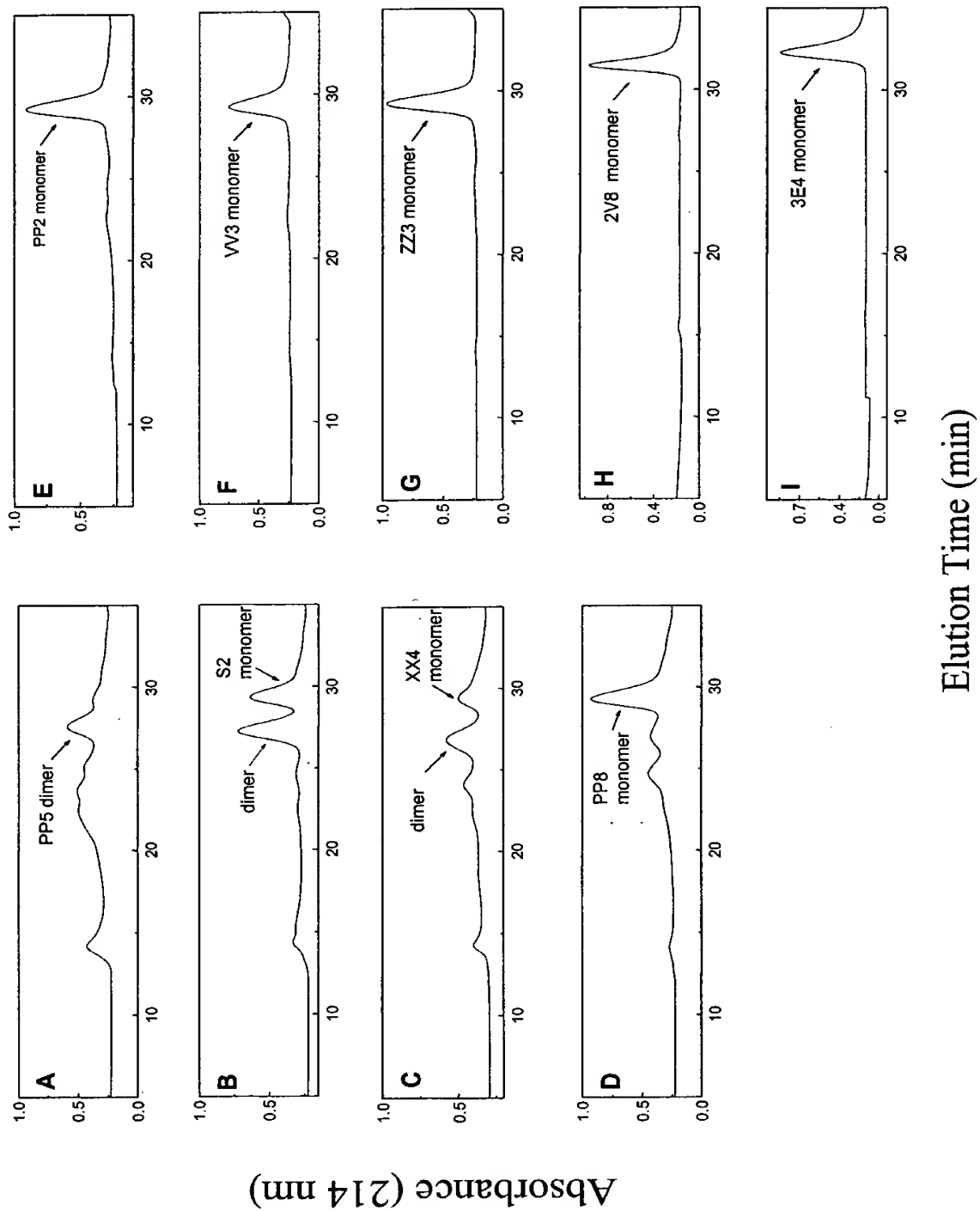
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	CTLA-4 VLD	(S2)	SPGKATE	YMMGNELTF	LMYPPPPYYL
A	CTLA-4-Som1	(PP2)	AGCKNFFWKFTFTSC	YMMGNELTF	LMYPPPPYYL
B	CTLA-4-Som1-tag	(PP5)	AGCKNFFWKFTFTSC	YMMGNELTF	LMYPPPPYYL
C	CTLA-4-Som3	(PP8)	SPGKATE	YMMGNELTF	AGCKNFFWKFTFTSC
D	CTLA-4-HA2	(XX4)	SPGKATE	YPYDVDPYA	LMYPPPPYYL
E	CTLA-4-Som1-Som3	(VV3)	AGCKNFFWKFTFTSC	YMMGNELTF	AGCKNFFWKFTFTSC
F	CTLA-4-Som1-HA2-Som3	(ZZ3)	AGCKNFFWKFTFTSC	YPYDVDPYA	AGCKNFFWKFTFTSC
G	CTLA-4-Anti-Lys	(2V8)	SGYTIGPYCMG	TYMMGNELTF	DSTIYASYECGHGLSTGGYGYDS
H	CTLA-4-Anti-Mel	(3E4)	SGFTFFSSYAMS	AISGSGGSTY	GWGLRGEEDYYMDV
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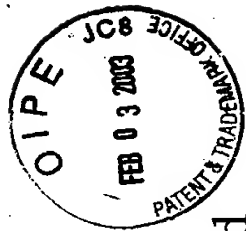
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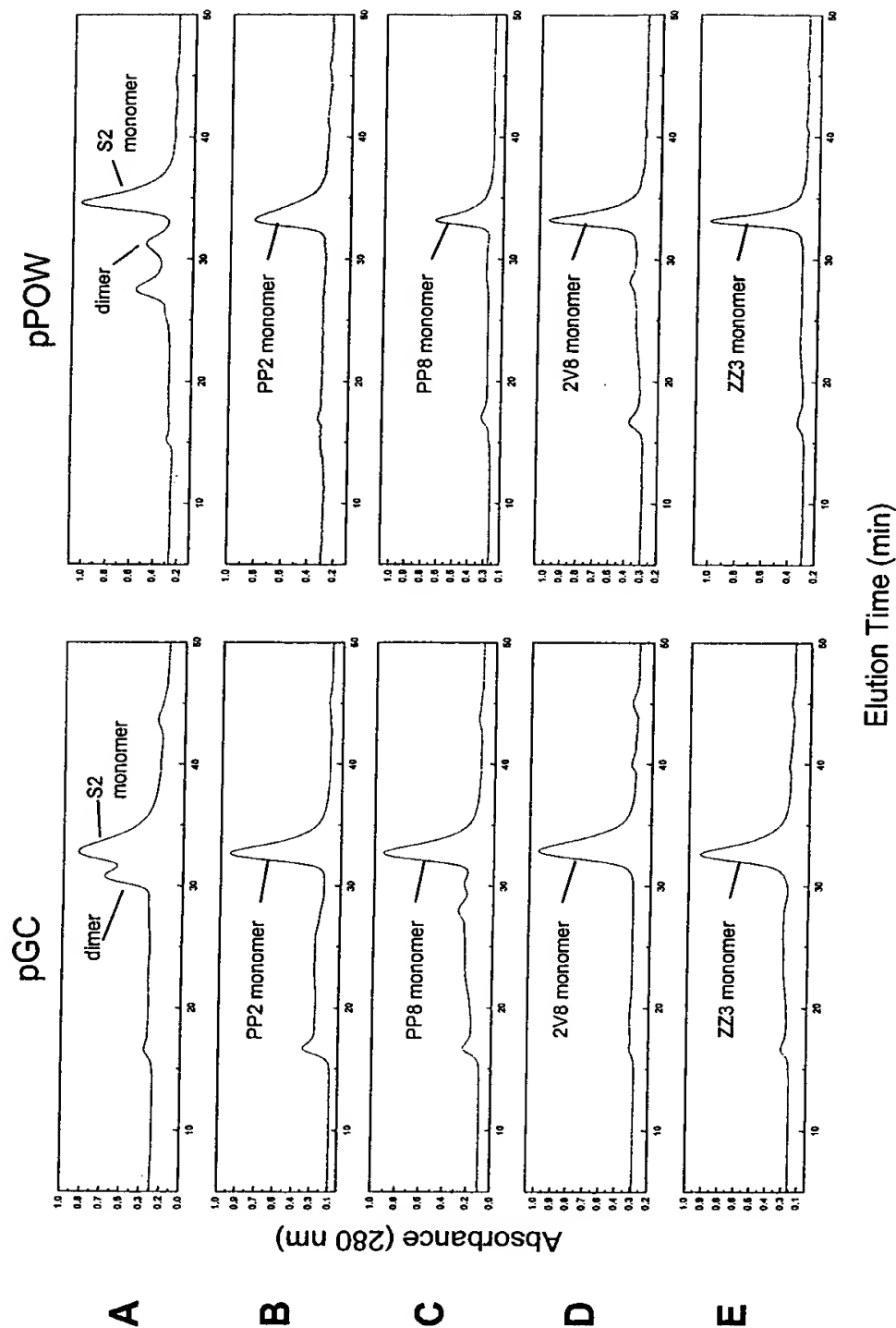
Figure 7: HPLC profiles of affinity purified recombinant CTLA-4 STMs

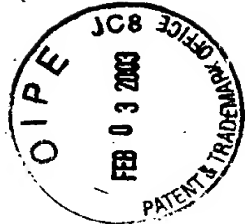


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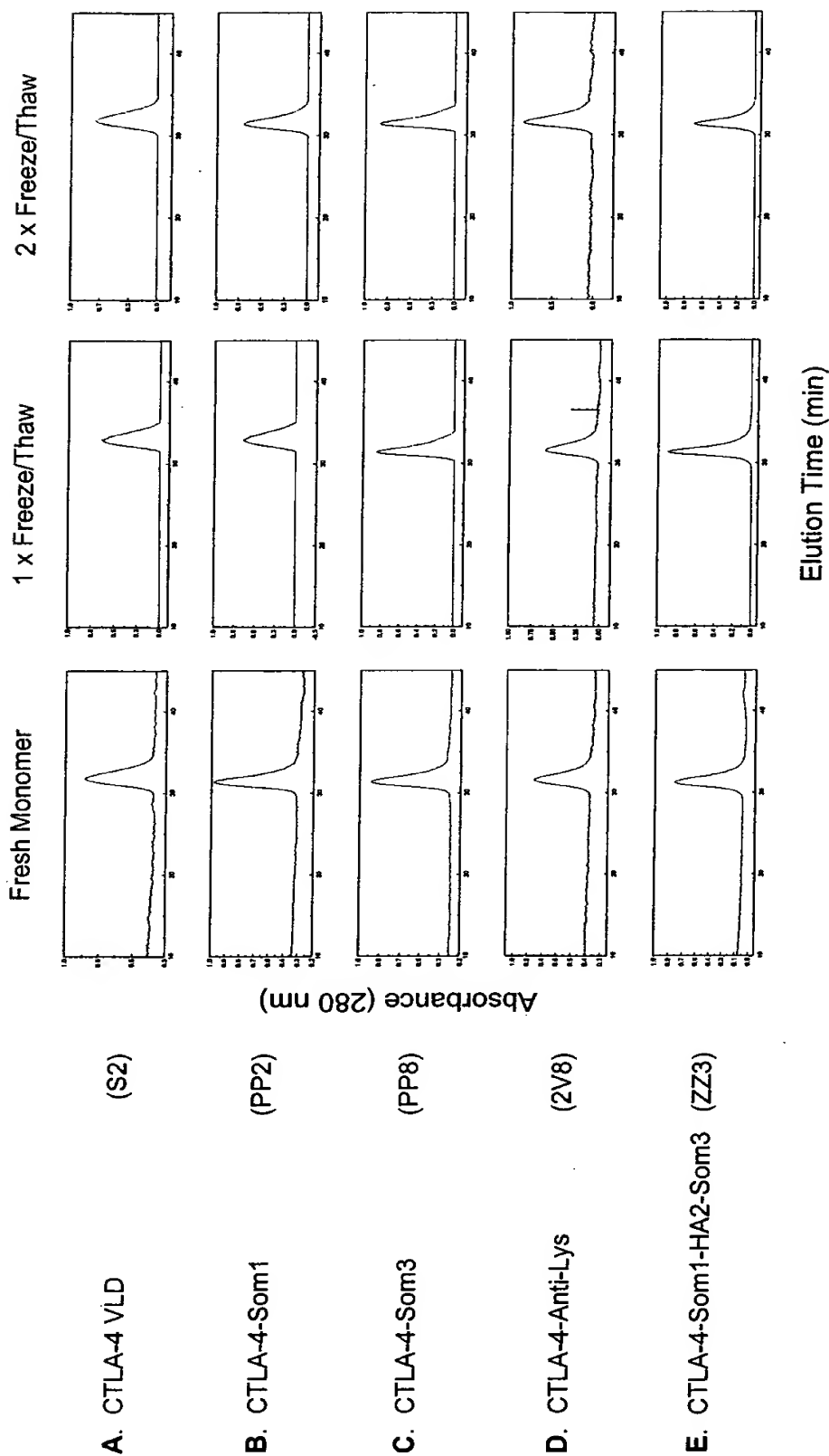
**Figure 8:** Comparison by size exclusion HPLC analysis of affinity purified CTLA-4 VLD STM constructs synthesised using bacterial expression vector pGC or pPOW.





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Figure 9: Size exclusion HPLC analysis of affinity purified CTLA-4 VLD STMs: Effect of Freeze/Thaw upon protein



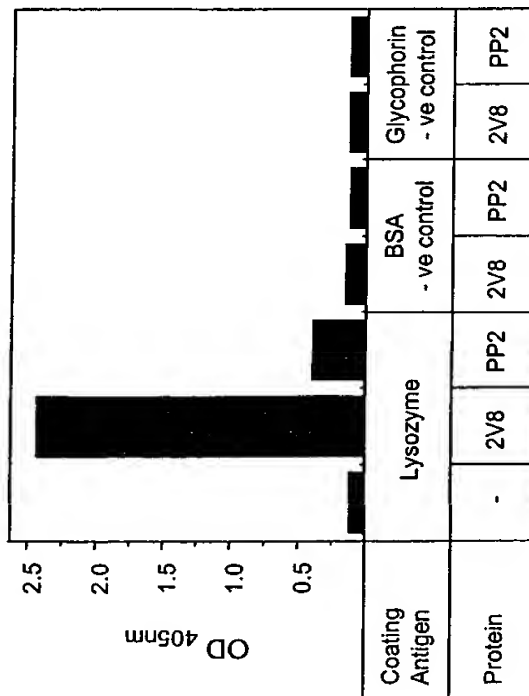
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Figure 10: Lysozyme binding characteristics of CTLA-4 anti-lysozyme construct 2V8

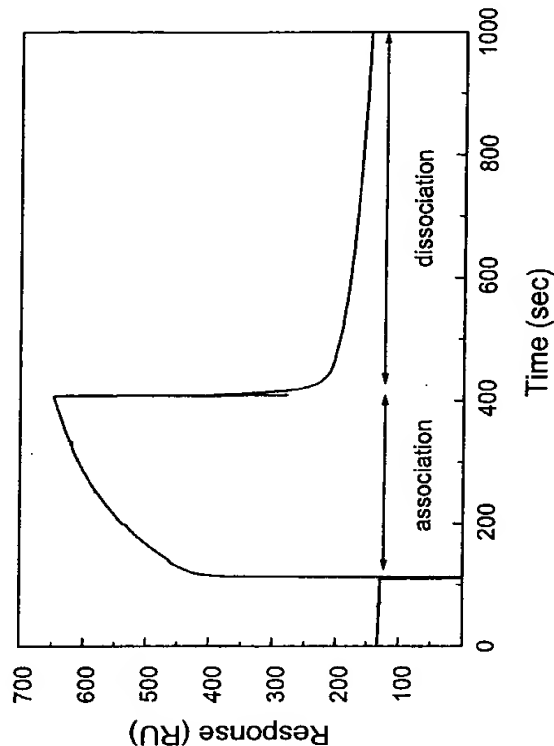
**A**

ELISA analysis of binding of 2V8 and PP2 constructs to lysozyme



**B**

BIAcore analysis of binding of lysozyme to 2V8 captured on immobilised anti-FLAG antibody



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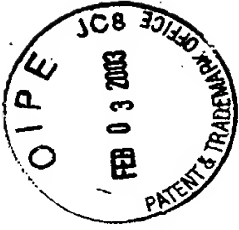
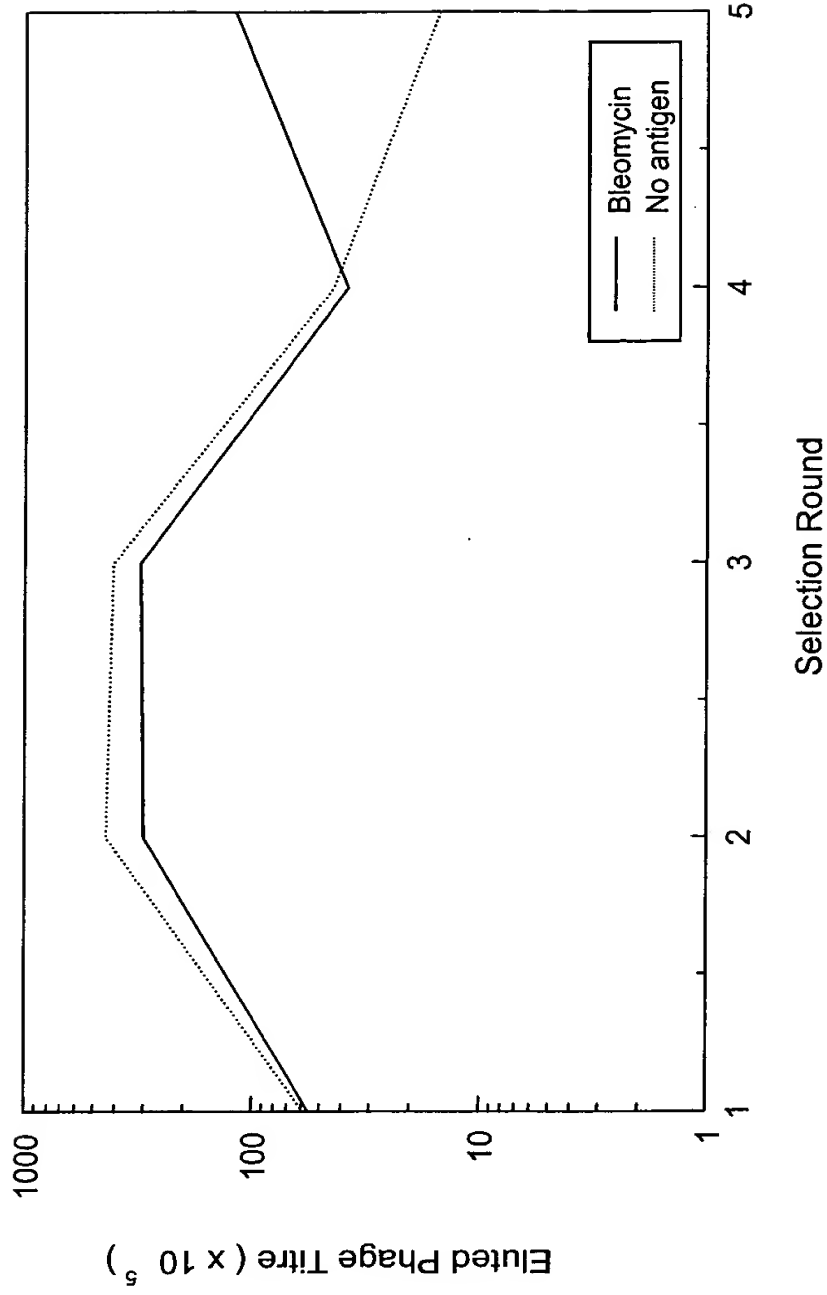


Figure 11: Screening of CTLA-4 VLD Phagemid Library on Immobilised Sh Bleomycin





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Figure 12: Screening of CTLA-4 VLD libraries in solution.

